

consists of a single step extraction by guanidinium isothiocyanate and phenol - chloroform.

26-37 KW 6/11/09
On pages 34-35, please replace paragraph [0118] with the following:

[0118] Phosphorothioate antisense oligonucleotides may be used (39). Modifications of the phosphodiester linkage as well as of the heterocycle or the sugar may provide an increase in efficiency. With respect to modification of the phosphodiester linkage, ~~phosphorothioate-phosphorothioate~~ may be used. An N3'-P5' phosphoramidate linkage has been described as stabilizing oligonucleotides to nucleases and increasing the binding to RNA (40). Peptide nucleic acid (PNA) linkage is a complete replacement of the ribose and phosphodiester backbone and is stable to nucleases, increases the binding affinity to RNA, and does not allow cleavage by RNase H. Its basic structure is also amenable to modifications that may allow its optimization as an antisense component. With respect to modifications of the heterocycle, certain heterocycle modifications have proven to augment antisense effects without interfering with RNase H activity. An example of such modification is C-5 thiazole modification. Finally, modification of the sugar may also be considered. 2'-O-propyl and 2'-methoxyethoxy ribose modifications stabilize oligonucleotides to nucleases in cell culture and in vivo. Cell culture and in vivo tumor experiments using these types of oligonucleotides targeted to c-raf-1 resulted in enhanced potency. As general references for antisense oligonucleotides, see (32-34)

On pages 45 and 46, please replace paragraphs [0144] and [0145] with the following: